Introgression of Wild Species Germplasm with Extreme Resistance to Cold Sweetening into the Cultivated Potato

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ABSTRACT

Potato (*Solanum tuberosum* L.) breeders are interested in developing chipping cultivars that can be stored at cold temperatures to reduce storage losses and increase profitability for potato producers. Commercial cultivars accumulate reducing sugars during cold storage, resulting in unacceptably dark chips when processed. In this study, we have identified diploid wild *Solanum* species accessions that are resistant to cold-induced sweetening at very low storage temperatures (2°C). Selected accessions were crossed as males to haploids (2*n* = 2*x*) of *S. tuberosum* to produce adapted hybrids, some of which produce acceptable chips following 3 mo of storage at 2°C. Reconditioning for 6 d at 20 to 22°C increased the number of clones with acceptable chip scores by threefold. The best wild species parents were *S. raphanifolium* 296126, 310998, and 210048. While parental chip scores help to predict offspring performance, progeny testing is important to identify the best cross combinations. The best hybrids have been introgressed into diploid and tetraploid breeding clones. These hybrids produce good tuber type and low levels of reducing sugars under extremely low storage temperatures.

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Abbreviations: chc, *Solanum chacoense*; oka, *Solanum okadae*; QTL, quantitative trait locus; rap, *Solanum raphanifolium*; spl, *Solanum sparsipilum*; stn, Stenotonum Group.

Processing potatoes (*Solanum tuberosum* L.) are an important value-added commodity. Over half of the U.S. potato crop is used for processing, and 20% of processed potatoes are made into chips, with annual sales over $6 billion (Anonymous, 2007). Because of the value of potato chips and their short shelf life (4–6 wk), potato processors require a constant, high-quality raw supply of chipping potatoes throughout the year (Brewer et al., 1990; Thill and Peloquin, 1994). Potatoes harvested in the spring and summer are rarely stored before processing, but potatoes harvested in the fall are stored for up to 7 mo before being processed (Hayes and Thill, 2003). One of the most important quality factors is chip color. Consumers demand light-colored chips because dark chips are unattractive and have an undesirable flavor (Burton, 1969; Coffin et al., 1987).

Chipping potatoes are typically held at storage temperatures of about 10°C (Blenkinsop et al., 2004). When stored below 10°C, they accumulate undesirable levels of the reducing sugars fructose and glucose and are said to undergo cold-induced sweetening (Blenkinsop et al., 2004; Denny and Thornton, 1940; Gould et al., 1979). When fried, they produce dark-colored chips because reducing sugars interact with amino acids in the nonenzymatic Maillard reaction (Denny and Thornton, 1941; Habib

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and Brown, 1957). The negative relationship between high reducing sugar content and light chip color is well documented (Sweetman, 1930).

Reducing sugar content in stored potatoes is influenced by cultivar genotype (Coffin et al., 1987; Gould et al., 1979). Storage and production environments also affect chip color. Chip color deteriorates with increasing amounts of time in cold storage (Stevenson and Cunningham, 1961) and with reductions in storage temperature (Gould et al., 1979). Even a single degree change in storage temperature affects reducing sugar content (Samotus et al., 1974). Environmental variables during the growing season, such as air temperature, soil temperature, moisture availability, and soil type, affect reducing sugar content and chip color (Sowokinos et al., 1987). Tuber maturity at harvest can also affect sugar content and chip color. As tubers become more mature with later harvest dates, reducing sugar levels decrease (Hope et al., 1960). In addition, immature tubers deteriorate in quality faster than mature tubers during cold storage (Walkof and Chubey, 1969). During the preconditioning period immediately after harvest, immature potatoes require more time to optimize sugar levels before being placed into cold storage (Herman et al., 1995).

Reconditioning (controlled tuber warming) after cold storage decreases levels of reducing sugars and is a common practice when potatoes are stored, especially below 10°C (Accatino, 1973; Habib and Brown, 1957). Storage buildings are warmed to 21 to 27°C for several weeks before shipment to processors. Potatoes subjected to warmer temperatures following cold storage metabolize reducing sugars, causing their concentrations to decline. The length of the reconditioning period depends on reducing sugar concentrations and their rates of decline. Not all genotypes respond similarly to reconditioning, however, and some do not respond at all (Stevenson and Cunningham, 1961).

Although chipping potatoes are typically stored at 7 to 13°C, colder storage temperatures are desirable (Thill and Peloquin, 1995). Storage temperatures of 4 to 6°C are the most favorable for long-term storage because losses due to rot, disease, and respiration are reduced at cold temperatures (Sowokinos, 2001a). Cold storage temperatures also reduce winter heating costs and the need for chemical treatments in storage to control disease and sprouting. Recent regulatory changes in the United States have reduced allowable residue levels of sprout inhibitors on potato tubers, generating more interest in the use of cold storage temperatures to control sprouting (Anderson et al., 2005).

Breeders are interested in the development of chipping cultivars with resistance to cold sweetening directly from cold storage or after a minimal reconditioning time (Hayes and Thill, 2002c; MacKay et al., 1990; Periera et al., 1994; Xiong et al., 2002). In the United States, recently released cultivars that produce light colored chips at storage temperatures of 5.6 to 7.2°C include 'Norvalley', 'Gemchip', 'MaineChip', 'Snowden', and 'White Pearl'. Some Japanese cultivars and breeding clones have low levels of reducing sugar content following storage at 4°C (Matsuura-Endo et al., 2004). Other breeding clones can also chip directly out of 3.3 to 4.5°C storage temperatures (Accatino, 1973; Anderson et al., 2005; Domanski et al., 2004; Hayes and Thill, 2002d; Lauer and Shaw, 1970; McKenzie et al., 2005; Oltmans and Novy, 2002). These clones contain wild and cultivated *Solanum* species in their pedigrees. These successes have led others to investigate exotic germplasm as new sources of resistance to cold sweetening (Jakuczun and Zimnoch-Guzowska, 2004; Thill and Peloquin, 1994).

Chip color is a heritable trait, with heritability estimates reported to be 0.81 to 0.87 following cold storage (Cunningham and Stevenson, 1963) and 0.86 following reconditioning (Accatino, 1973). Additive effects were found to be important for the control of glucose content in cold-stored diploid clones (Jakuczun and Zimnoch-Guzowska, 2004). Resistance to cold sweetening may be controlled by only a few genes. Two-locus (Accatino, 1973; Lynch et al., 2003) and three-locus (Thill and Peloquin, 1994) models have been hypothesized for resistance to cold sweetening, while a three-locus model has been proposed for reconditioning ability (Thill and Peloquin, 1994). These models generally require dominant alleles at all loci for the production of light-colored chips. Recessive genes with epistatic interactions may also influence resistance to cold sweetening (Colon et al., 1989). Thirteen genetic markers, representing six quantitative trait loci (QTLs), have been found to influence chip color (Douches and Freyre, 1994). These six QTLs explained 43.5% of the phenotypic variation in a segregating population. When one significant epistatic interaction was included, 50.5% of the variation was explained. An additional study found 24 QTLs for glucose, fructose, and sucrose content distributed along all 12 potato chromosomes (Menendez et al., 2002). A candidate gene approach, based on a molecular function map for carbohydrate synthesis and transport (Chen et al., 2001), was used to correlate allelic variation with resistance to cold sweetening. Associations between alleles of the carbohydrate metabolism genes invertase (Li et al., 2005)and UDP-glucose pyrophosphorylase (Sowokinos, 2001b) have been reported.

Progress in the development of cultivars with resistance to cold sweetening requires the creation and identification of appropriate parents. When parents of varying chip quality are crossed, clones that produce light-colored chips following reconditioning may be found in all families, but the highest frequencies result when both parents produce light-colored chips (Cunningham and Stevenson, 1963; Ehlenfeldt et al., 1990). High midparent progeny correlations (0.84–0.92) indicate that mean parent chipping performance can predict progeny performance. Other researchers have also noted that the use of parents with good chipping quality increases the frequency of progeny with light chip

color (Accatino, 1973; Ehlenfeldt et al., 1990; Pereira et al., 1993; Thill and Peloquin, 1994).

Solanum tuberosum germplasm is not considered a good source of genes for resistance to cold sweetening (Pandey et al., 2005). On the other hand, wild diploid *Solanum* species are valuable sources of these genes (Hanneman, 1993, 1996; Hayes and Thill, 2002a). Wild species, however, do not tuberize under the long day conditions of the north temperate regions (Jansky et al., 1990; Rudorf, 1958). It is difficult to evaluate them for tuber traits such as resistance to cold sweetening unless tubers are generated in a greenhouse. When wild species are crossed to haploids ($2n = 2x = 24$) derived from tetraploid *S. tuberosum* ($2n = 4x = 48$) cultivars, the resulting haploid-species hybrids are often adapted and can be evaluated for tuber traits (Hermundstad and Peloquin, 1986).

This study focuses on the use of wild *Solanum* species with resistance to sweetening at colder temperatures than in previous studies (2°C). These species were crossed as males to *S. tuberosum* haploids to create adapted hybrids for chip evaluation. The goal of the research was to develop haploid × species hybrids that would produce light-colored chips directly following a 3-mo storage period at 2°C or after a short reconditioning period of 6 d at 20 to 22°C.

MATERIALS AND METHODS

Experiment 1 versus Experiment 2

Two experiments are reported in the study. Experiment 1 includes an initial screen of wild species and a limited set of crosses made to utilize those species. The first year of the study (1994) was not replicated and served as an initial evaluation of hybrids between selected wild species accessions and a few cultivated female parents. In 1995, families in this experiment were replicated. Experiment 2 was performed in 2 yr (1995 and 1996) and expanded on Exp. 1 by utilizing additional wild and cultivated parents, and by generating a larger set of families. Families in Exp. 2 were replicated in both years.

Experimental Design

In Year 1 of Exp. 2, families were replicated by planting half of the seedlings into one replication and half into the second replication. In Year 2 of Exp. 1 and 2, families were replicated by planting half of the tubers from a family bag into one replication and the remaining tubers into the second replication. Unless otherwise indicated, a randomized complete block design with two replications was used. All studies were performed at the University of Wisconsin Lelah Starks Potato Breeding Farm, Rhinelander, WI. In-row spacing was 60 cm, and between-row spacing was 90 cm. Standard cultural practices were used throughout the growing season. Check cultivars included 'Atlantic', 'Katahdin', 'Kennebec', 'Langlade', 'Snowden', 'Superior', and 'Wischip'.

Chip Quality Evaluation

For all experiments, direct chipping evaluations were performed on tubers stored at 2°C for 3 mo. The reconditioning treatment was performed for 6 d at 20 to 22°C. Each tuber was evaluated for chip color by taking a 1- to 2-mm slice from the center of a transverse cut, rinsing it in tap water, and frying it in 190°C corn oil until bubbling ceased. Each chip was visually scored for color using a scale of 1.0 to 10.0 (dark), at 0.5 intervals, based on the International Chip Institute (Cleveland, OH) color chart.

Experiment 1: Parents

Seven accessions from four wild diploid *Solanum* species were selected for use as male parents based on a high frequency of plants with acceptable direct chip scores (accession mean ≤ 4.5) in two prior years of evaluation (Table 1). They were *S. okadae* (oka) 498063; *S. raphanifolium* (rap) 296126, 310998, and 458384; *S. sparsipilum* (spl) 458386 and 473385; and *S. tarijense* 473336.

Five *S. tuberosum* haploids ($2n = 2x = 24$) derived from four tetraploids were selected as female parents based on high female fertility and early maturity. A haploid–*S. chacoense* (chc) hybrid was also chosen as a parent, based on light chip color after a 16-d reconditioning period at 20 to 22°C. One selected haploid clone (US-W730) had a reconditioned chip score of 7, four clones (US-W1, US-W457, US-W973, and US-W13125) had scores of 8, and the haploid–chc hybrid had a score of 4.5. Female parents had scores of 10 when they were chipped from cold storage without reconditioning.

Experiment 1: Hybrid Families

In 1994, 22 haploid × species hybrid families were created in a greenhouse in Madison, WI. Pollen from 10 to 25 plants per wild species accession was bulked and used in pollinations with the six selected female parents. Seeds were extracted, soaked in 4 mM gibberellic acid for 24 h, and then sown in a greenhouse at Rhinelander. Hybrid families were transplanted to peat pots 2 wk later and transplanted to the field along with check cultivars, without replication, 6 wk after that (11–12 July). Hybrid families ranged in size from 16 to 195 plants. Vines were mechanically removed 1 wk before harvest, and tubers were machine harvested on 29 September. All plants that tuberized were collected as families. One tuber from each plant was collected in a family bag. One family bag was collected for direct chipping, one for reconditioning, and a third for planting the following year. Data were tabulated on a family basis.

In 1995, the seven check cultivars and the 22 hybrid seedling tuber families, ranging in size from 10 to 90 clones, were planted

Table 1. Chip score means and ranges of wild Solanum species accessions selected as male parents for Exp. 1. Data are averaged across 2 yr. Tubers were stored for 3 mo at 2°C.

Species	Accession	Direct chip score mean†	No. plants < 4.5‡ (% of total)
S. okadae	498063	5.3	18 (50)
S. raphanifolium	296126	4.6	19 (50)
S. raphanifolium	310998	5.5	17 (49)
S. raphanifolium	458384	6.4	20 (35)
S. sparsipilum	458386	7.3	9 (16)
S. sparsipilum	473385	6.7	9 (24)
S. tarijense	473336	4.5	11 (100)

†Chip scale from 1 to 10 (dark).

‡An acceptable chip score is <4.5.

on 31 May. Vines were mechanically removed 1 wk before harvest. Plants were machine harvested on 12 September. Family bags were collected for chip evaluation as previously described.

Experiment 2: Parents

Thirty-one accessions representing 12 wild diploid *Solanum* species were selected as male parents based on 3 yr of direct chipping data (Table 2). Accessions were categorized by direct chip color means as being good (≤ 6.0), medium (6.1–8.0), or poor (>8.0). All accessions were also reconditioned for 6 d at 20 to 22°C and scored for chip color (Table 2).

Female parents included one *S. tuberosum* Andigena Group haploid and 18 *S. tuberosum* Tuberosum Group haploids extracted from four tetraploid cultivars, one Wisconsin breeding line, and one Minnesota breeding line. Haploids were selected for female fertility and/or chipping ability after reconditioning. Reconditioned chip scores were based on 1993 field-grown tubers. Female clones were considered good (≤6.0), medium (6.1–8.0), or poor (>8.0) for chipping based on mean chip scores after reconditioning in 1993 and 1994. Four haploids were considered good, eight medium, and seven poor clones. All clones scored 9 or 10 when chipped directly out of cold storage.

Table 2. Chip score means and ranges of wild *Solanum* species accessions selected as male parents for Exp. 2. Data are averaged across 3 yr for direct chip scores and based on 1 yr for the reconditioned score. Tubers were stored for 3 mo at 2°C.

Species	Accession	Direct chip score mean†	Reconditioned chip score mean‡
S. capsicibiccatum	473458	6.1 (2–10)	7.1 (4–10)
S. gourlayi	472995	9.8 (8–10)	7.8 (2–10)
S. gourlayi	472999	8.8 (4–10)	7.9 (4–10)
S. medians	283081	5.8 (3–10)	5.1 (3–9)
S. medians	310994	6.8 (3–10)	3.9 (3–9)
S. okadae	498063	5.8 (3–10)	6.0 (3–10)
S. okadae	498064	5.9 (3–10)	5.3 (3–9)
S. okadae	498065	8.1 (3–10)	5.4 (3–10)
S. okadae	498130	7.3 (3–10)	6.7 (3–10)
S. tuberosum Phureja Group	225673	8.2 (3–10)	8.3 (5–10)
S. tuberosum Phureja Group	243465	9.3 (5–10)	7.3 (5–10)
S. raphanifolium	210048	6.2 (3–10)	6.2 (4–10)
S. raphanifolium	265862	6.3 (3–10)	6.1 (2–10)
S. raphanifolium	296126	5.9 (3–10)	6.4 (3–10)
S. raphanifolium	310998	4.3 (3–9)	5.5 (4–10)
S. raphanifolium	458384	7.2 (3–10)	6.8 (3–10)
S. raphanifolium	458408	4.5 (4–10)	6.7 (4–9)
S. raphanifolium	473371	5.7 (3–9)	5.8 (3–7)
S. raphanifolium	473466	5.8 (3–10)	8.3 (7–10)
S. raphanifolium	473467	4.9 (4–9)	4.0 (3–8)
S. sanctae-rosae	320325	8.2 (4–10)	9.8 (9–10)
S. sogarandinum	365360	5.8 (3–10)	5.3 (3–10)
S. sparsipilum	458386	7.6 (4–10)	7.0 (3–10)
S. sparsipilum	473385	7.5 (3–10)	6.9 (3–10)
S. sparsipilum	498137	8.0 (3–10)	7.4 (3–10)
S. sparsipilum	498305	7.6 (3–10)	6.8 (3–10)
S. tuberosum Stenotomum Group	230512	8.6 (5–10)	7.8 (6–10)
S. tuberosum Stenotomum Group	205527	7.0 (4–10)	7.4 (4–10)
S. tarijense	473232	8.7 (5–10)	8.1 (4–10)
S. tarijense	473238	7.7 (4–10)	7.3 (4–10)
S. verrucosum	275250	6.9 (3–10)	5.4 (3–10)

†Chip scale from 1 to 10 (dark).

‡Reconditioned for 6 d at 20 to 22°C.

Experiment 2: Hybrid Families

The 19 selected haploids were pollinated with bulked wild species pollen (5–100 plants per accession) to create 158 hybrid families. All parents were represented in at least one hybrid family. In 1995, seedling families containing up to 180 plants were transplanted to the field along with tubers of the seven check varieties on 20–21 June. Vines were mechanically removed before harvest and tubers were machine harvested on 25–27 September. One tuber from each plant that tuberized was collected in a family bag. One family bag was collected for a direct chipping test, one for reconditioning, and a third for planting in 1996.

Tuber families harvested in 1995 were planted on 7 June 1996, vine killed on 9 September, and harvested on 19 September. One tuber from each plant that tuberized was collected in a family bag. One family bag was collected for a direct chipping test, one for reconditioning, and a third for maintenance.

Data Analysis

For both experiments, progeny within each family were grouped into three chip score categories, good (≤4.5), medium (mean score of 5–7), and poor (>7). Analyses of variance using the General Linear Model in SAS (SAS Institute, Cary, NC) were performed on families containing at least 15 clones. Regression analysis was performed between midparent values and progeny means to estimate narrow sense heritability for chip score. Correlation coefficients were calculated to compare chip scores between years and to compare direct chip scores with reconditioned chip scores.

RESULTS

Direct Chip Evaluations, Experiment 1

The 22 hybrid families produced 1622 seedlings, of which 889 (55%) tuberized in 1994. In 1995, 726 seedling tubers were planted, of which 404 (56%) tuberized. In 1994 and 1995, 15/889 (1.7%) and 11/404 (2.7%) clones, respectively, produced acceptable direct chip scores (≤4.5). Family means for direct chip scores ranged from 7.4 to 10.0 in 1994 and from 6.4 to 9.9 in 1995. In both years, significant variation ($P < 0.01$) was detected among families. Family H25 [(US-W973 × chc) × rap 296126] ranked first for direct chip color in 1994, while H25 and H28 [(US-W973 ×

chc) × rap 310998] ranked first in 1995. Most families in which rap 296126 or rap 310998 were the male parents and US-W973 × chc was the female parent ranked high, while families involving rap 458384 ranked low.

Comparisons between years revealed that chip color in each family except H10 (US-W457 × spl 458386) was lighter in 1995 than in 1994. Three families, H10, H8 (US-W457 × rap 458384), and H15 (US-W730 × spl 458386), produced only poor chipping progeny in both years. All check varieties had direct chip scores of 10 in both years. The ANOVA of the 13 families with at least 15 progeny in both years indicated significant variation between years ($P < 0.01$), among families ($P < 0.01$), and for the year × family interaction ($P = 0.01$). The correlation coefficient between years for direct chip score based on family means was $r = 0.78$ ($P < 0.01$).

When all progeny with good chip scores were pooled by male parent, the highest percentages of good scores (≤4.5) and medium scores (5–7) were produced by rap 296126 and rap 310998. They accounted for all progeny with good scores in 1994 and 90.9% in 1995. They were also parents of 87.8% of progeny with medium chip scores in 1994 and 75.4% in 1995. In 1995, rap 458384 was the parent of a single clone with an acceptable chip score. Families from these three rap accessions, each crossed to the same female parents, were compared for chipping performance. There were typically no significant differences between family means involving rap 296126 and rap 310998, while both parents produced significantly better progeny than rap 458384 ($P = 0.05$).

When pooled by female parent, US-W973 × chc produced the highest percentage of progeny with good chip scores. It accounted for 93.3% of all progeny with good chip scores in 1994 and 81.8% in 1995. The only other females that produced good direct chipping progeny were US-W973 in both years and US-W1 in 1995. Three sets of family comparisons were extracted to determine whether US-W973 × chc was a better parent than other females when crossed to the same three rap accessions. In each comparison, families from US-W973 × chc were ranked first for mean chip color in both years.

Table 3. Direct chip scores of adapted germplasm. Tubers were stored for 3 mo at 2°C (2003–2004) or 4.4°C (2005–2007).

Parentage†	Ploidy	2007‡	2006	2005	2004	2003
(H28 × H28) × W1351	4*x*	3	4.5	4.5	5	9
(H25 × W870) × W870	4*x*	5	5.5	5	9	10
Yukon Gold × AH60-20	4*x*	6	6	5	7	10
(H25 × W870) × W1005	4*x*	6.5	7.5	4.5	8	9
White Pearl × (H25-9 × W1005)	4*x*	5.5	5.5	4.5	4	6
White Pearl × (H25-9 × W1005)	4*x*	5	7	7	5	7
W1421 × (H25 × W870)	4*x*	4	4.5	5	5	6
H25-9 × W1005	4*x*	4	6	6	4	10
H25 [(US-W973 × chc) × rap 296126]	2*x*	3	4.5	3	3	5
H28-5 [(US-W973 × chc) × rap 310998]	2*x*	4	4	4	4	4
H28-6	2*x*	4	4	4	3	4.5
H28-7	2*x*	4.5	4	4.5	3	4.5
H28-10	2*x*	5.5	4.5	4	4	4.5
(US-W5536.7 × 8030.8) × H28-18	2*x*	4	5	4.5	4	4.5
H28 × H28	2*x*	3	6	3	3	3
[(US-W973 × chc) × rap 310998] × full-sib	2*x*	3.5	4.5	4	3	4
US-W357 × phu 225673	2*x*	4.5	6	5	5	4.5
US-W357 × rap 458384	2*x*	7	4.5	3	4	3
AH60-1 (US-W357 × tar 473238)	2*x*	4	4	3	4	4.5
AH66-51	2*x*	5	6	4	4	4
AH72-2 (US-W3694 × stn 230512)	2*x*	4	6	5.5	4	5
US-W10349 × ver 275250	2*x*	6	4	4	4	4.5
US-W10349 × med 283081	2*x*	4.5	3	3	4	6
(US-W2836 × US-W5314.3) × (H28 × H28)	2*x*	4	5	4	3	4
[(H28–18 × chc) × (US-W751 × US-W42)] × (H28 × H28)	2*x*	3	4	3	4	4.5
(US-W3694 × phu 225673) × AH60-20	2*x*	6	8	4.5	4	3
(US-W3694 × phu 225673) × (H28 × H28)	2*x*	3.5	3	4.5	4	3
(US-W4056 × phu 225673) × AH60-1	2*x*	3.5	3	3	4	4
(US-W4056 × stn 205527) × AH60-20	2*x*	5.5	4	3	4	4.5
(US-W4056 × stn 205527) × (H28 × H28)	2*x*	3.5	5	5.5	4	3
[(H28 x H28) × (US-W9523.21 × US-W9591.4)] × AH60	2*x*	3	3	3	4	4
[(H28 x H28) × (US-W9523.21 × US-W9591.4)] × AH60-1	2*x*	5	3	3	4	3
(US-W4056 × stn 205527) × AH60-20	2*x*	4.5	5	4	8	8[3]
(US-W4056 × stn 205527) × AH60-20	2*x*	3	4	4	8	9[3]
(US-W4056 × stn 205527) × AH60-20	2*x*	3	4	7	7	7.5[§]
(US-W4056 × stn 205527) × [(US-W5536.7 × 8030.8) × H28-18]	2*x*	3	4.5	4	7	6.5[§]
(US-W4056 × stn 205527) × [(US-W5536.7 × 8030.8) × H28-18]	2*x*	4.5	5	4.5	7	7.5[§]
(US-W4056 × stn 205527) × [(US-W5536.7 × 8030.8) × H28-18]	2*x*	4.5	4	3	8	5[3]

†W numbers = advanced selections from the Wisconsin breeding program; US-W numbers = haploids; phu = Phureja Group; rap = *S. raphanifolium*; ver = *S. verrucosum*; med = *S. medians*; chc = *S. chacoense*; tar = *S. tarijense*; stn = Stenotomum Group.

‡Chip scale from 1 to 10 (dark).

§Stored at 3.3°C.

Direct Chip Evaluations, Experiment 2

In 1995, 3472 progeny from 158 families were chipped directly out of cold storage. Fifty families produced 92 progeny with direct chip scores of 4.5 or lower, accounting for 2.6% of the total. In 1996, 1217 progeny from 61 families were chipped directly out of storage. Nineteen families produced 31 clones with direct chip scores of 4.5 or lower, accounting for 2.5% of the total. All check cultivars scored 9.5 or 10 in both years. Most progeny scores were in the 7 to 10 range (Fig. 1). There was significant variation among families ($P < 0.01$) and for the replication × family interaction ($P < 0.01$) in 1995 and 1996. Replications were significantly different from each other in 1995 ($P = 0.01$), but not in 1996 ($P = 0.14$).

When data were pooled by wild species parent, the best in both years was rap 210048. It produced 17.4 and 22.6% of the progeny with direct chip scores of 4.5 or lower in 1995 and 1996, respectively. Interestingly, it was classified as a medium chipping accession, with a chip color mean of 6.2. *Solanum okadae* 498063 was also a good parent, producing 12.0 and 12.9% of the progeny with direct chip scores of 4.5 or lower in 1995 and 1996, respectively. The best wild species accessions for producing offspring with direct chip scores in the medium range (5–7) were rap 210048, rap 458384, rap 296126, and *S. tuberosum* Phureja Group 225673.

When data were pooled by haploid parent, the three haploids that produced high percentages of good progeny were US-W10349 (17.4 and 9.8%), US-W357 (16.3 and 25.8%) and US-W3694 (16.3 and 19.4%) in 1995 and 1996, respectively. All of these haploids were derived from the cultivar Merrimack. They were scored as good clones for chipping following reconditioning, but their direct chip scores were poor (9 or 10).

Table 4. Reconditioned chip scores of adapted germplasm. Tubers were stored for 3 mo at 2°C (2003–2004) or 4.4°C (2005–2007) and reconditioned for 2 wk at 20–22°C.

Parentage†	Ploidy	2007‡	2006	2005	2004	2003
(H28 × H28) × W1351	4*x*	3	5.5	4	4.5	7
(H25 × W870) × W870	4*x*	4	4	4	5	4.5
Yukon Gold × AH60-20	4*x*	5	6	4	6	8
(H25 × W870) × W1005	4*x*	6	5.5	7	4	8
White Pearl × (H25-9 × W1005)	4*x*	3	4.5	4.5	4	6
White Pearl × (H25–9 × W1005)	4*x*	4	5	4	4.5	6
W1421 × (H25 × W870)	4*x*	3.5	3	4	4.5	4.5
H25-9 × W1005	4*x*	4.5	6	4	3	8
H25 [(US-W973 × chc) × rap 296126]	2*x*	3	4.5	3	6	–
H28-5 [(US-W973 × chc) × rap 310998]	2*x*	3.5	4	4	5	4
H28-6	2*x*	4	4	4	6	3
H28-7	2*x*	4	4	5	7	4
H28-10	2*x*	6.5	4.5	5	5	–
(US-W5536.7 × 8030.8) × H28-18	2*x*	3	5	3	6	4
H28 × H28	2*x*	4	6	4	7	–
[(US-W973 × chc) × rap 310998] × full-sib	2*x*	3.5	4.5	4	7	–
US-W357 × phu 225673	2*x*	–	6	7	8	10
US-W357 × rap 458384	2*x*	3	4.5	7	9	10
AH60-1 (US-W357 × tar 473238)	2*x*	3	4	5	9	7
AH66-51	2*x*	5	6	4	8	8
AH72-2 (US-W3694 × stn 230512)	2*x*	4	6	3	9	9
US-W10349 × ver 275250	2*x*	4	4	3	10	9
US-W10349 × med 283081	2*x*	6.5	3	3	5.5	8
(US-W2836 × US-W5314.3) × (H28 × H28)	2*x*	4	5	4	8	6
[(H28–18 × chc) × (US-W751 × US-W42)] × (H28 × H28)	2*x*	3.5	4	4.5	4.5	3.5
(US-W3694 × phu 225673) × AH60-20	2*x*	4	8	6	10	10
(US-W3694 × phu 225673) × (H28 × H28)	2*x*	3.5	3	3	6	5
(US-W4056 × phu 225673) × AH60-1	2*x*	3	3	4	7	5
(US-W4056 × stn 205527) × AH60-20	2*x*	3.5	4	4	7	7
(US-W4056 × stn 205527) × (H28 × H28)	2*x*	3	5	4	4.5	5
[(H28 × H28) × (US-W9523.21 × US-W9591.4)] × AH60	2*x*	3	3	4	7	4.5
[(H28 × H28) × (US-W9523.21 × US-W9591.4)] × AH60-1	2*x*	3	3	4	5	4
(US-W4056 × stn 205527) × AH60-20	2*x*	3	4.5	4	4.5	4[3]
(US-W4056 × stn 205527) × AH60-20	2*x*	3	3	3	3	6§
(US-W4056 × stn 205527) × AH60-20	2*x*	3	3	4	3	4§
(US-W4056 × stn 205527) × [(US-W5536.7 × 8030.8) × H28-18]	2*x*	3	4	4	4	3§
(US-W4056 × stn 205527) × [(US-W5536.7 × 8030.8) × H28-18]	2*x*	4.5	5	3	4	5§
(US-W4056 × stn 205527) × [(US-W5536.7 × 8030.8) × H28-18]	2*x*	4	4	4	5	4§

†W numbers = advanced selections from the Wisconsin breeding program, US-W numbers = haploids, phu = Phureja Group, rap = *S. raphanifolium,* tar = *S. tarijense*; ver = *S. verrucosum*, med = *S. medians*, chc = *S. chacoense,* stn = Stenotomum Group.

‡Chip scale from 1 to 10 (dark).

§Stored at 3.3°C.

Reconditioned Chip Evaluations, Experiment 1

Reconditioned chip scores of 4.5 or less were observed for 50/691 (7.2%) and 37/367 (10.1%) of the progeny in 1994 and 1995, respectively. Family means of reconditioned chip scores ranged from 6.0 to 9.7 in 1994 and from 5.6 to 8.8 in 1995. In each year, ANOVA detected significant variation ($P < 0.01$) among families. Family H25 [(US-W973 × chc) × rap 296126] ranked first in both years, as it was in the direct chip studies.

As with the direct chip study, all families except for H10 (US-W457 × spl 458386) had a lighter chip color in 1995 than in 1994. Among the seven cultivars, Snowden had the lightest reconditioned chip color in both years (6.5 and 5.0 in 1994 and 1995, respectively). An ANOVA of the 11 families with at least 15 progeny in both years indicated significant variation between years ($P < 0.01$) and among families ($P < 0.01$), but not for the year × family interaction ($P = 0.39$). The correlation coefficient between years based on family means was $r = 0.64$ ($P < 0.01$).

All progeny with good reconditioned chip scores (≤4.5) were pooled by male parent. The highest percentages of progeny with good scores were produced by rap 310998 and rap 296126. *Solanum raphanifolium* 310998 ranked first in both years, producing 44.0 and 35.1% of the good progeny in 1994 and 1995, respectively. *Solanum raphanifolium* 296126 ranked second in both years, producing 32.0 and 27.0% of the good progeny in 1994 and 1995, respectively, while rap 458384 ranked third. Families from these three rap accessions, each crossed to the same set of female parents, were compared for reconditioned chip performance. As with the direct chip study there was no significant difference between means of families from rap 296126 and rap 310998, but mean chip scores of these two families were significantly better than those involving rap 458384 ($P = 0.05$).

When pooled by female parent, US-W973 × chc produced a higher percentage of progeny with good reconditioned chip scores than any other female in both years. It produced 80.0 and 48.6% of all progeny with good reconditioned chip scores in 1994 and 1995, respectively. Haploid US-W730 ranked second, producing the next highest percentages of progeny with good reconditioned chip scores in both years. Three sets of family comparisons were extracted from all families to determine whether the US-W973 × chc female was a better parent than other females when crossed to the same three rap accessions. Families produced by US-W973 × chc had the lightest reconditioned chip color in both years ($P = 0.05$).

Reconditioned Chip Evaluations, Experiment 2

In 1995, 2890 progeny from 148 families were chipped after reconditioning. Of those families, 88 produced 412 progeny (14.3%) with good reconditioned chip scores (≤4.5) (Fig. 2). Among the cultivar checks, Snowden had the lightest reconditioned chip score in both years (5 and

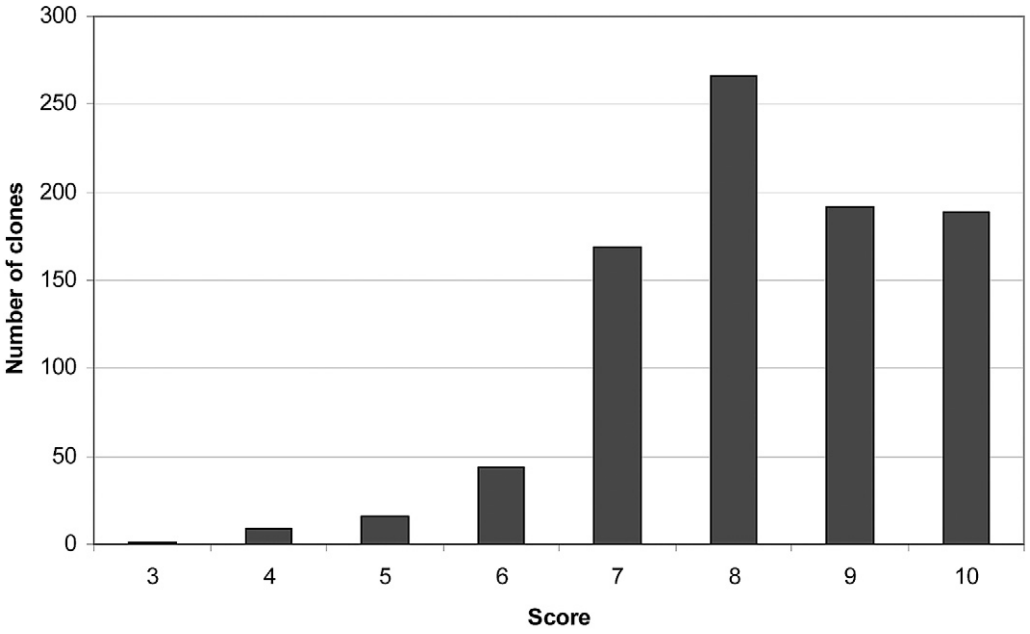

Figure 1. Frequency distribution of direct chip scores of all clones in Exp. 2, 1996. An acceptable chip score is ≤4.5.

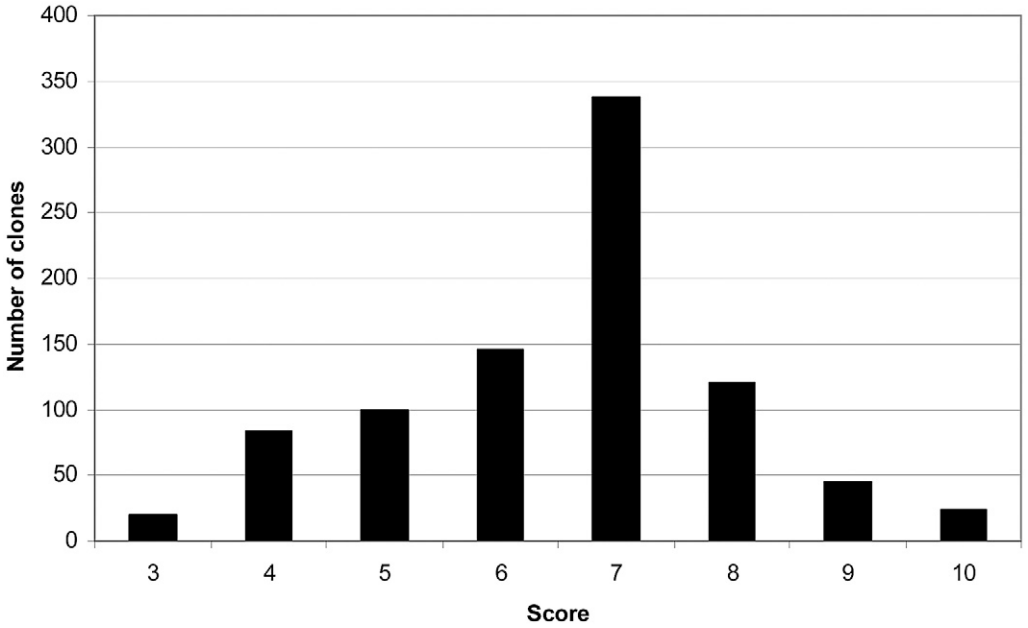

Figure 2. Frequency distribution of reconditioned chip scores of all clones in Exp. 2, 1996. An acceptable chip score is ≤4.5.

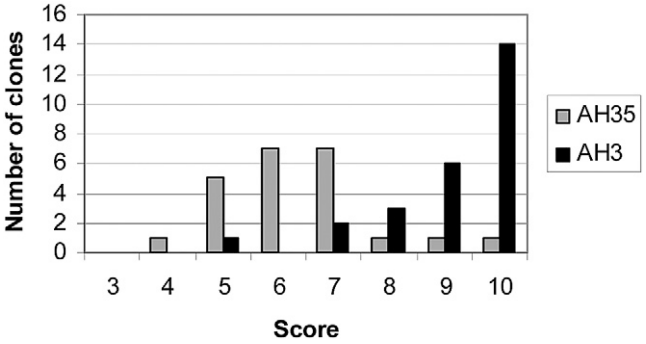

Figure 3. Frequency distribution of direct chip scores of the families with the highest and lowest mean direct chip scores in Exp. 2, 1996. AH35 = US-W4212 × *Solanum raphanifolium* 310998; AH3 = US-W527 × Phureja Group 225673. An acceptable chip score is ≤4.5.

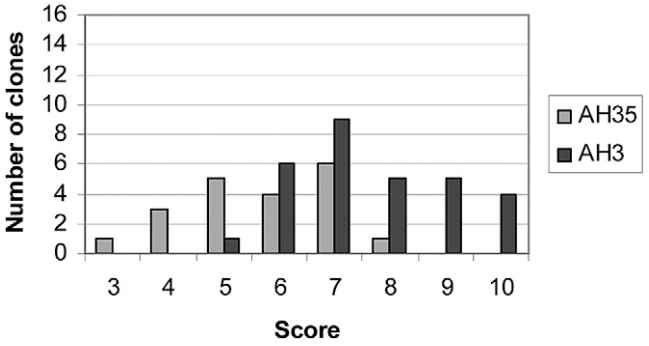

Figure 4. Frequency distribution of reconditioned chip scores of the families with the highest and lowest mean direct chip scores in experiment 2, 1996. AH35 = US-W4212 × *Solanum raphanifolium* 310998; AH3 = US-W527 × Phureja Group 225673. An acceptable chip score is ≤4.5.

5.5 in 1995 and 1996, respectively). In 1996, 1151 progeny from 62 families were chipped after reconditioning. Of these families, 31 produced 124 progeny (10.8%) with good reconditioned chip scores. Compared to chip scores directly out of cold storage, a shift occurred in the distribution of scores toward lower values (Fig. 1 and 2).

Significant differences in reconditioned chip scores were detected among families ($P < 0.01$) and between replications ($P < 0.01$) in both years. The replication × family interaction was not significant in either year ($P = 0.36$ and 0.06 in 1995 and 1996, respectively).

When data were combined by wild species parent, rap 210048 produced the highest percentage of progeny with good reconditioned scores (12.1% in 1995 and 23.6% in 1996). Other accessions that produced a high percentage of progeny with good reconditioned chip scores in both years were oka 498063 (9.5 and 8.0%) and rap 473371 (9.2 and 7.3%). Wild species parents that produced a high percentage of offspring with medium reconditioned chip scores (5–7) include *S. tuberosum* Stenotonum Group (stn) 230512 and rap 458384.

Considering data pooled by haploid parent, US-W357 accounted for the highest percentage of progeny with good reconditioned chip scores (37.1 and 61.0% in 1995 and 1996, respectively). It was scored as a clone with good reconditioning ability. Other haploid parents that produced a high proportion of progeny with good reconditioned chip scores include US-W4056 (10.7 and 8.1%) and US-W3694 (9.7 and 6.5%). While US-W4056 produced medium chip scores following reconditioning, US-W3694 produced good scores.

Direct versus Reconditioned Chip Scores

In both years, reconditioning improved chip scores. In Exp. 1 in 1994, mean chip color was improved in all families after reconditioning. In 1995, it improved in all families except H9 (US-W457 × rap 310998) and H14 (US-W730 × rap 296126). In 1994 and 1995, 35/691 (5.1%) and 26/367 (7.1%) more progeny had good chip scores (≤4.5) after reconditioning. Based on family means, correlation coefficients between direct and reconditioned chip scores were $r = 0.78$ ($P < 0.01$) and $r = 0.74$ ($P < 0.01$) in 1994 and 1995, respectively.

In Exp. 2 in 1995, all family means improved after reconditioning except for AH116 (US-W4056 × spl 498305) and AH92 (US-W3817 × stn 230512). In 1996, all family means except AH 219 (US-W551 × rap 458384) improved after reconditioning. A total of 92/3472 (2.6%) and 31/1217 (2.5%) clones chipped in the good category directly out of cold storage, while 412/2890 (14.3%) and 123/1151 (10.7%) clones were good after reconditioning in 1995 and 1996, respectively. After reconditioning, a majority (54.0 and 62.1%) of the progeny chipped in the medium category in 1995 and 1996, respectively. The correlation coefficient between direct chipping and reconditioning scores was $r = 0.59$ ($P < 0.01$) and $r = 0.63$ ($P < 0.01$), in 1995 and 1996, respectively.

Midparent Progeny Regressions

Based on direct chip scores in Exp. 1, midparent progeny regression values were $R^2 = 0.24$ ($P = 0.03$) and $R^2 = 0.45$ ($P = 0.01$) in 1994 and 1995, respectively. In Exp. 2, they were $R^2 = 0.38$ ($P = 0.01$) and $R^2 = 0.36$ ($P < 0.01$). Based on reconditioned chip scores, midparent progeny regression values in Exp. 1 were $R^2 = 0.57$ ($P < 0.01$) and $R^2 = 0.42$ ($P < 0.01$) in 1994 and 1995, respectively. In Exp. 2, they were $R^2 = 0.33$ ($P < 0.01$) and $R^2 = 0.39$ ($P < 0.01$) in 1995 and 1996, respectively.

DISCUSSION

Direct Chip Scores of Families

We accomplished the primary goal of this research by developing adapted haploid × wild species clones with resistance to cold sweetening at very low temperatures. These clones are useful as parents for the development of cultivars with exceptional resistance to cold sweetening. As expected, most progeny and all check cultivars chipped in the poor category at a 2°C storage temperature. This is an extreme storage temperature and has not been used in published efforts to breed for resistance to cold sweetening. Previous studies have been performed at 3 to 4°C, but the

ability to store at 2°C is likely to profoundly improve the ability to store tubers for a long period of time with minimal losses. The relatively small temperature drop from 4 to 2°C places extreme physiological demands on tubers, resulting in poor chip quality unless clones with exceptional resistance to cold sweetening are identified. About 2% of the progeny screened in this study produced light-colored chips directly out of 2°C storage. At 4.4°C, 5.4% of a diploid population produced light-colored chips when processed directly from storage (Lauer and Shaw, 1970); at 10°C, 39.4% of another diploid population produced light-colored chips (Thill and Peloquin, 1994). While each of these studies used different populations, a common trend is apparent. As storage temperature decreases, it becomes increasingly difficult to identify clones with resistance to cold sweetening. If the potatoes in this study were stored at warmer temperatures, their reducing sugar levels would likely have been lower (Denny and Thornton, 1941; Gould et al., 1979), resulting in a larger pool of clones with good chip scores.

The germplasm used for this study consisted of haploid × wild species hybrids. While wild species do not tuberize in the field, more than half of the hybrids produced enough tubers for chip evaluations. When a haploid × chc clone was used as a female parent in crosses to wild species, percentage of tuberization in offspring was reduced compared with families in which the female parent was a haploid (45 vs. 63% in 1994; 51 vs. 58% in 1995). As expected, within a family, the proportion of clones that tuberize in the field decreases as the amount of wild species germplasm in the pedigree increases.

Most families exhibited large variation for chip color, with chip scores among family members ranging from low to high. The range of variation for direct and reconditioned chip color of two families is represented in Fig. 3 and 4, respectively. It is not surprising that the families in this study were highly variable for chip color. The parents are self-incompatible and presumably highly heterozygous. The wild *Solanum* species contribution is expected to be especially heterogeneous, since bulk pollen samples consisted of multiple plants within an accession. Intra-accession variation is common in wild *Solanum* species, resulting in the need for the fine screening of accessions (Bamberg et al., 1996; Douches et al., 2001; Jansky et al., 2006, 2008; Zlesak and Thill, 2004). In this study, pollen was bulked to survey the range of genotypes in an accession. In future work, it would be desirable to use fine screening to select individual wild species clones for use as parents. This strategy will likely produce a larger proportion of progeny with resistance to sweetening at very low temperatures.

Effect of Parent on Progeny Direct Chip Scores

In Exp. 1, families H25 [(US-W973 × chc) × rap 296126] and H28 [(US-W973 × chc) × rap 310998] were exceptional in both years. These families were generated from the same female parent crossed to different accessions of rap. Even lower family mean scores were observed in Exp. 2, with an Andigena haploid crossed to oka 498063 and Merrimack haploids crossed with additional accessions of rap (458408 and 296126). The larger set of parental combinations in Exp. 2 produced a wider range of progeny means at both ends of the spectrum, resulting in the identification of additional clones with lighter chips directly from 2°C storage. One clone from family H28 (H28-6) and one from family AH66 (AH66-1) have subsequently been found to consistently produce a light chip color (<2.0 on a scale of 1–5) directly out of 3°C storage (Oltmans and Novy, 2002).

Direct chip scores of wild species parents were closely related to offspring performance. In both years of Exp. 1, rap 296126 and rap 310998 produced more progeny with good chip scores than any other accession. Together they accounted for 100% of all good progeny in 1994 and 90.9% in 1995. Accession 296126 was reported by Oltmans and Novy (2002) to have exceptional chip color directly from 2°C storage. In our initial screening study, these two accessions were ranked among the best three direct chipping accessions, with mean scores of 6.0 or less. In Exp. 2, rap 210048 produced the highest percentage of the total good direct chipping progeny even though its mean chip score was 6.2. *Solanum okadae* 498063 and rap 310998, with mean chip scores of 5.8 and 4.3, respectively, produced the next highest percentages of good progeny. Accessions with low mean chip scores produced a higher percentage of good progeny than any other group of accessions. Other researchers have also observed that using parents with good chip scores or low reducing sugar levels increases the frequency of progeny with good chip scores (Accatino, 1973; Cunningham and Stevenson, 1963; Pereira et al., 1993; Thill and Peloquin, 1994).

Clones with medium or even poor chip scores may hold some potential as parents. When a medium chipping parent, such as rap 210048, was used in Exp. 2, progeny with good chip scores were produced. Similarly, in Exp. 2, a high percentage of good direct chipping progeny was produced by US-W13030, even though the parent itself had a direct chip score of 10. These genotypes must contain favorable alleles for direct chip quality even though they do not express them. Others have reported similar results, in which crosses between poor and good direct chipping parents produced some progeny with good direct chip scores (Ehlenfeldt et al., 1990; Thill, 1994). The three-gene model of Thill and Peloquin (1994) can explain how parents with poor chip color could produce progeny with light chip color. A dominant allele at each of three loci (A_/B_/C_) is needed to express the trait. Consequently, the genotype AA/BB/cc is considered poor for chipping because only two loci are dominant. Crossing this clone to a parent with a good chip score, such as Aa/Bb/CC, would produce progeny with light-colored chips.

Reconditioning scores of haploids may provide the best tool for predicting their potential contribution to chip quality of their progeny directly from cold storage. All haploid parents scored 9 or 10 when chipped directly from 2°C storage. However, they did vary in chip color following reconditioning. In Exp. 2, haploids US-W10349, US-W357, and US-W3694 produced dark-colored chips directly from cold storage, but reconditioning resulted in lighter chips. These clones produced the highest percentages of good direct chipping progeny. Thill and Peloquin (1994) reported higher frequencies of progeny with good direct chip scores from 10°C storage when both parents had good chip scores following reconditioning (40.7%) than when good × poor (38.8%) and poor × poor (37.4%) crosses were made.

The value of exotic germplasm was apparent even among the female parents. In Exp. 1, US-W973 × chc was the best female clone for chip scores following reconditioning. When chipped directly from cold storage, its offspring were superior to those produced by the haploid female parents even when they had the same wild species parents. The chc parent of this clone probably provided favorable alleles for chip quality directly out of cold storage. When both US-W973 × chc and US-W973 were crossed to the same rap accessions (296126 and 458384), families from US-W973 × chc (H25 and H26) were ranked higher than families from US-W973 (H1 and H2) for direct chip color score.

It is difficult to consistently predict the best parental combinations under these extreme storage temperatures solely on the basis of the direct chipping performance of parents. Midparent values were influenced only by wild species performance because there was no variation in female direct chip scores. Although midparent progeny regression analyses revealed significant associations, they were relatively small in both experiments, ranging from $R^2 = 0.24$ to 0.45. Similarly, Thill (1994) did not find a clear relationship between parent and offspring when tubers were chipped directly from 4°C storage. It would be prudent to carry out progeny tests using small populations when making decisions about parents for the development of populations with resistance to cold sweetening.

Reconditioned Chip Scores of Families

We also produced adapted hybrids with the ability to generate acceptable chips after a short reconditioning period. In most cases, chips from reconditioned tubers were lighter than those from tubers taken directly out of cold storage. In Exp. 1, the short reconditioning period increased the number of good chipping progeny more than threefold, from 15/889 (1.7%) to 50/691 (7.2%) in 1994 and 11/404 (2.7%) to 37/367 (10.1%) in 1995. A similar increase was noted in Exp. 2, from 92/3472 (2.6%) to 412/2890 (14.3%) in 1995 and from 31/1217 (2.5%) to 124/1151 (10.8%) in 1996. Tubers held at warmer temperatures after cold storage begin to metabolize reducing sugars, resulting in a decline in their levels. The length of reconditioning time required for the production of acceptable chips depends on genotype, storage temperature, and the length of the storage period before reconditioning. While most clones improved after reconditioning, some quite dramatically, others did not respond to reconditioning. Thill (1994) also noted that most clones produced better chip color, but some produced poorer chip color after reconditioning. Stevenson and Cunningham (1961) found that clones varied in the number of weeks needed to produce acceptable chips during reconditioning. However, some clones did not recondition acceptably even after 7 wk.

A 6-d reconditioning period is extremely short for material stored at 2°C for 3 mo, but we took this approach to identify exceptional parents for resistance to cold sweetening. Typically, cold-stored potatoes are warmed to 21 to 27°C for several weeks before reducing sugar levels are acceptable for processing. However, we identified several clones that reconditioned after a short period at room temperature. Accatino (1973) also identified germplasm that chipped acceptably after a short reconditioning period following storage at 3.3°C. The identification of parents that produce acceptable progeny following a short reconditioning period may offer a compromise to direct chipping. This strategy provides more clones from which to select for other important traits, such as dry matter content and disease resistance. In addition, a brief reconditioning period would provide a desirable alternative to a long reconditioning period because it is less expensive and less time consuming for growers.

Effect of Parent on Progeny Reconditioned Chip Scores

As with direct chipping, it is difficult to predict superior parents for the production of progeny with good chip scores following reconditioning. Many clones, including some with poor or medium chip scores, were among the parents of the top-ranking families. However, 100% of the wild species accessions with good direct chip scores and 61.5% of the haploid clones with good chip scores following reconditioning were parents of families ranked in the top 50%. Others have found that crosses between the best parents tend to produce the best families for chip color after reconditioning, but crosses involving clones with less-than-good chip scores can also produce acceptable offspring (Accatino, 1973; Cunningham and Stevenson, 1963; Pereira et al., 1993). It is important to note that progeny with good chip quality can be produced by parents that are not among the best for chip color following reconditioning. For example, in Exp. 2, rap 210048 produced the highest percentage of progeny with good chip quality following reconditioning even though it was classified as a medium reconditioning accession. From a breeding perspective, parents with moderate chip scores may possess other desirable traits that are not found in the best chipping

clones, so that overall, they may also make valuable contributions to the development of superior cultivars.

All wild species accessions produced some offspring with good chip scores. However, in Exp. 1, rap 296126 and rap 310998 produced the highest percentages of progeny with good chip scores after reconditioning in both years. Combined, they accounted for 76.0 and 62.1% of the good progeny in 1994 and 1995, respectively. Although wild species parents in Exp. 1 were not reconditioned, these two accessions ranked among the top three for direct chip scores. This again supports a breeding strategy based on the concept that better chipping parents give rise to a higher frequency of progeny with light chip color.

In contrast to the direct chipping studies, the reconditioning performance of progeny can be predicted, in part, on the basis of the reconditioning ability of the female parents. In Exp. 1, US-W973 × chc produced a higher percentage of progeny with good reconditioned chip scores than any other female in both years. Haploid US-W730 ranked second, producing the next highest percentages of progeny with low reconditioned chip scores in both years. In Exp. 2, good and medium reconditioning haploids produced better reconditioned offspring than poor reconditioning haploids. US-W357, the best reconditioning haploid, produced the highest percentages of progeny with chip scores following reconditioning. US-W973 and US-W13030 were also good parents. However, US-W13030 produced progeny with good chip scores even though its reconditioned chips were dark. Apparently, it has favorable alleles for reconditioning but does not express them. Thill (1994) also noted that one of the parents in his study produced a high frequency of acceptable reconditioning progeny even though it was a poor reconditioning parent.

As with the direct chip studies, midparent progeny regression values for chip color following reconditioning were significant, but moderate, ranging from $R^2 = 0.33$ to 0.57. This indicates that additive genetic variance is not a major component of the total phenotypic variance. Consequently, progeny testing is important to identify superior parents. Thill (1994) found no consistent pattern between the midparent mean and the family mean. However, a higher midparent progeny regression value ($R^2 = 0.69$) was observed in a different population (Accatino, 1973), perhaps reflecting different sources of genes that contribute to cold sweetening resistance.

Comparison between Direct and Reconditioned Chip Scores

Significant and positive correlations were detected between direct chip scores and those following reconditioning. In Exp. 1, correlation coefficients were $r = 0.78$ ($P < 0.01$) in 1994 and $r = 0.74$ ($P < 0.01$) in 1995. The correlation coefficient in Exp. 2 was similar (0.59, $P < 0.01$). Other researchers have noted positive correlations between chip color directly from cold storage and after reconditioning, with values of 0.74 to 0.80 (Loiselle et al., 1990), 0.73 (Thill, 1994), and 0.81 to 0.94 (Accatino, 1973). These correlations indicate that the genes controlling direct chip color may be linked to those that control chip color after reconditioning (Accatino, 1973; Thill and Peloquin, 1994). Alternatively, there may be an overlap in the two sets of genes.

Effect of Environment on Chip Score

Based on family means in Exp. 1, chip score data appear to be consistent between the 2 yr of the study. The correlation coefficient for direct chip data of families was $r = 0.78$ ($P < 0.01$), while that for reconditioned chip data was $r = 0.64$ ($P < 0.01$). High correlation coefficients for reconditioned families across years have been previously reported by Cunningham and Stevenson (1963) ($r = 0.90$) and Accatino (1973) ($r = 0.78$).

High correlation coefficients between replications in both studies indicate consistency across environments within a production year. In Exp. 1, there was no significant effect of replication or the replication × family interaction. However, significant variation due to replications and the replication × family interaction was detected in Exp. 2. Others have noted signification variation for direct chip color due to replication, the genotype × replication interaction, and the location × genotype interaction (Scheffer et al., 1992) and location, the year × genotype interaction, and the location × genotype interaction (Thill, 1994).

Reducing sugar levels are affected by the production environment and tuber maturity. Environmental stresses such as heat, moisture, and soil temperature affect chip color and/or sugar content of potatoes during growth and storage (Motes and Greig, 1970; Sowokinos et al., 1987). Differences in growing conditions may have affected chip color in both experiments. Although meteorological data were not collected, 1994 was cooler, it had more rainfall, and the trial was planted in a heavier soil type than in 1995. Overall, 1995 was a better chipping year as indicated by means of families and check varieties. Heavier soils are cooler because they hold more moisture. Cooler soil temperatures reduce potato tuber respiration rates, which slows the breakdown of starch and sugars, ultimately resulting in darker-colored chips due to high sugar levels. Similarly, wet summers produce darker-colored chips. High soil temperatures have also been associated with dark chip color (Motes and Greig, 1970). Moisture and heat stress experienced by the plant during the growing season may cause tubers to form regions with high reducing sugars during storage (Sowokinos et al., 1987). In contrast, moderate daytime temperatures and low levels of precipitation result in tubers with low levels of reducing sugars. The light chip colors observed in 1995 may have resulted from these conditions.

Physiological differences between tubers generated in the 2 yr of each experiment may account for another source

of variation in chip color. In Year 1 of Exp. 1, tubers were chipped from transplant families originating from true seed, while in Year 2, they were chipped from seedling tuber families originating from the previous year's transplants. The transplants had a shorter growing season than did plants grown from seedling tubers. Tubers from transplant families did not have the opportunity to mature as much as did those grown from tubers. Tuber maturity has an effect on sugar development and concentration (Burton, 1969; Hope et al., 1960; Nelson and Sowokinos, 1983; Weaver and Timm, 1983). Reducing sugar content decreases with successively later harvest dates when clones are planted at the same time. Immature tubers have a high sugar content and deteriorate in quality faster than mature tubers (Herman et al., 1995; Nelson and Sowokinos, 1983; Walkof and Chubey, 1969).

Because the goal of this research was to introgress genes for resistance to cold sweetening into adapted germplasm, we followed up on Exp. 1 and 2 by crossing selected clones with diploid breeding lines to improve adaptation and tuber quality. In addition, some selected diploid clones were crossed to tetraploid cultivars and breeding lines to produce adapted tetraploid clones via unilateral sexual polyploidization (Chase, 1963). This breeding scheme has been used by others to transfer resistance to cold sweetening from diploid to tetraploid breeding clones (Hayes and Thill, 2002b). The tetraploid progeny we have produced by unilateral sexual polyploidization are directly crossable to cultivars in breeding programs. A list of these clones and their chip scores across years is presented in Table 3 (direct) and Table 4 (reconditioned). It is apparent from these data and Exp. 1 and 2 that chip scores vary across years. In addition, some clones appear to be more stable than others. However, most clones with acceptable scores in one year have acceptable scores in other years. It is important to note that while many of these clones contain 25 to 50% wild species germplasm, they produce commercially acceptable tubers under the long days of temperate field production systems. Consequently, extensive backcrossing to cultivated germplasm is not necessary.

SUMMARY

Potato breeders place a high priority on the development of germplasm with resistance to cold sweetening. Many programs are focusing on the production of cold chipping cultivars because of the benefits of storing at cold temperatures. However, despite decades of effort to generate germplasm with resistance to cold sweetening, to our knowledge, this is the only published study in which wild *Solanum* species accessions with resistance to cold sweetening at very cold temperatures (2°C) have been introgressed into cultivated germplasm. Approximately 2% of progeny from crosses between cultivated and wild potato species produced acceptable chips after storage at 2°C for 3 mo. A short (6-d) reconditioning increased the proportion of acceptable progeny by threefold. Crosses of diploid wild species to haploids derived from the cultivated potato provided an effective method to introgress genes for resistance to cold sweetening into adapted germplasm. Subsequent crosses have produced diploid and tetraploid hybrids with good tuber yield and type while maintaining extreme resistance to cold sweetening.

The most effective predictions about direct chip scores of progeny can be made when considering the direct chip score of the wild species parent and the reconditioned chip score of the cultivated parent. Moderate midparent progeny regression values indicate that nonadditive genetic variance plays a major role in determining the chip phenotype following cold storage of tubers. Consequently, while it is helpful to choose parents on the basis of their chip scores, their true parental value cannot be determined without progeny testing. Strong correlations between direct and reconditioned chip scores indicate that selection for one is likely to lead to improvement for the other.

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